

NAI Teams Annual Project Report
2008 Annual Report
July 1, 2007 - June 30, 2008

Project Title

**DDF: Geomicrobiology of a Unique Ice-Sulfur Spring Ecosystem
in the High Canadian Arctic (Templeton, PI)**

Project Summary

A glacial, active sulfide spring environment at Borup Fiord Pass on Ellesmere Island in the Canadian High Arctic provides an excellent opportunity to study microbial life at a site that may be an analog to Europa, an ice-covered moon of Jupiter. During the past year we have collected samples from the extensive mats of sulfur-minerals that precipitate in discharge channels to identify the microbial communities hosted in the sulfur-ice, and to cultivate key organisms mediating the oxidation of H₂S to elemental sulfur.

Project Progress (Accomplishments)

In 2007-8, our team conducted integrated geological, geochemical and geomicrobiological surveys at a potential Europa analogue field site in the Canadian High Arctic where extensive sulfur biomineralization occurs on the ice-surface each spring. Our efforts focused on elucidating how the chemistry and geological structure of Borup Fiord Pass controls the metabolism and diversity of ice hosted microorganisms.

Summer field work was conducted by Steve Grasby and Benoit Beauchamp (Canadian Geological Survey) on northern Ellesmere Island. There was no active spring discharge at Borup Fiord at this time; however, recently formed discharge channels and extensive deposits of elemental sulfur were both observed. A profile of six sulfur-rich mineral samples were collected from the discharge point and downstream to the pro-glacial area for geochemical analysis, stable isotopic analysis, microbial culturing experiments and molecular phylogenetic characterization.

DNA was extracted from all mineral samples for analysis via 16/18S rRNA gene sequencing by John Spear and graduate student Chase Williamson at the Colorado School of Mines. Although bacterial and eucaryal sequences were identified using a variety of PCR primers, to date no archaeal sequences have been found. Based on DNA sequence similarity, bacterial diversity associated with the sulfur-rich springs at Borup Fiord Pass is similar to other cold, saline environments, including other polar springs and Antarctic environments. A number of phylotypes associated with sulfur metabolisms have been identified, such as sequences showing >98% BLAST identity with *Thiomicrospira arctica* and *Thiomicrospira pshychrophila*, chemolithoautotrophic, sulfur-oxidizing bacteria isolated from Arctic marine sediments (Knittel et al. 2005). Many sequences also correspond to *Sulfuricurvales* and *Sulfurovumales*, sulfur-oxidizing isolates recently detected in subglacial environments (Skidmore et al. 2005), and numerous lineages within the *Deltaproteobacteria*, which may represent ubiquitous sulfate-reducing organisms in this system. Initial analysis with the Ribosomal Database Project's Classifier software (Wang et al. 2007) also indicates that some of the 16S rRNA sequences from samples collected in 2007 may also be representative of a new Phylum of Bacteria.

All the mineral samples collected in 2007 were also used for the targeted culturing of psychrophilic microbial organisms involved in dark sulfur cycling by Alexis Templeton and graduate student Damhnait Gleeson. The primary objective was to obtain stable consortia of organisms mediating the oxidation of reduced sulfide and forming S^0 as a byproduct, using NaS “gradient tubes” to develop opposing fluxes of sulfide and oxygen. Optical microscopy revealed S^0 minerals associated with abundant extracellular material, forming sulfur-mineralized sheaths. After numerous successive rounds of transfers in which the S^0 appeared, DNA was then extracted from two stable consortia to identify the members present. All 16S rRNA sequences obtained from the consortia have been detected in the 2007 16S rRNA environmental clone libraries from Spear and Williamson, suggesting that the cultivated organisms are environmentally abundant and relevant. Using this information, dilution-to-extinction and serial plating techniques have been used to bring 80% of the organisms into pure culture, and currently all isolates have been reintroduced to sulfide gradient tubes to evaluate whether or not they can catalyze sulfide oxidation in pure culture.

Currently, all team members are integrating the geochemical, microbiological and molecular data to generate a series of publications to be led by graduate students Gleeson and Williamson (who presented their work at AbSciCon and the International Conference on Alpine and Polar Microbiology in Banff, Canada). First, sulfur stable isotopic data from the mineral samples needs to be analyzed in the context of the field geochemical data. Second, additional clone libraries are being processed to ensure deep coverage of microbial diversity. And lastly, full phylogenetic and physiological characterization of the sulfide-oxidizing isolates is being completed to determine specific metabolic roles in Borup Fiord microorganisms involved in producing S^0 as a potential biosignature.

Keywords

Sulfur, biosignature, Europa